Pyridine-2-Aldoxime Methiodide

A Valuable Agent for Phosphate Poisoning

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• **Phosphate insecticide use is increasing as is concomitant human poisoning. Home insecticide bomb as well as agricultural, crop contamination and suicidal exposure are noted.**

Clinical poisoning may be chronic and severe. It may follow long exposure or short exposure with heavy dosages. Manifestations are those of excessive cholinergic activity.

Adequate laboratory means for early, rapid diagnosis and screen testings are available.

PAM is a valuable agent for this type of poisoning and is a much more adequate and complete antidote than atropine. It is available (under certain restrictive conditions presently). It is being widely used elsewhere in the world but with limited education and use in this country. Morbidity and mortality continue at a rate that could probably be corrected.

Case reports, describing the use of this antidote in our hands, are included.

Government and industry responsibility as well as physician education must be more clearly defined in prevention, recognition and treatment in what is often a life threatening situation.

PHOSPHATE ESTER INSECTICIDES are being widely used in agriculture throughout the world. These compounds were initially developed as deadly weapons of twentieth century warfare in the form of nerve gases. They are now used for agricultural purposes in greater quantity than any other insecticide except D.D.T.[3] They are also used in the home, and residual deposits on food crops have caused poisoning of persons. Cases of poisoning from exposure of workers in greenhouses and from deliberate use as a suicidal agent have been reported.

They act by interfering with normal nerve impulse transmission. Acetylcholine is a chemical mediator of nerve impulses at the ganglionic and end organ sites of innervation. Acetylcholinesterase prevents excessive accumulation of acetylcholine by a hydrolytic process. These phosphate poisons form a covalent bond with acetylcholinesterase, resulting in an inactive phosphorylated substance which is no longer capable of hydrolysis of acetylcholine.[4] The acetylcholine thereby accumulates to abnormal degree, resulting in excessive parasympathomimetic effect.

CLINICAL POISONING

The incidence of clinical poisoning is difficult to determine. In 1957 in California, there were 229 cases of occupational organic phosphate insecticide poisoning reported, one of them fatal.[5]

It has been reported from Israel that Parathion leads the list of insecticides causing poisoning there and is one of the five major poisons causing fatalities. Many suicides from Parathion have been reported in Finland. Food contamination poisoning has been reported, as have numerous instances of accidental ingestion by children. The greatest incidence, however, lies in exposure of personnel involved in many phases of agriculture, such as crop dusting, handling of plants and greenhouse work. In one case a farmer died of poisoning that resulted from walking through a field that had been dusted with one of these phosphate compounds. He had taken no part in the dusting process.

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A bureau of the California State Department of Occupational Health has established regulations regarding protective clothing and equipment to prevent undue exposure. Also defined are time intervals during which persons should avoid dusted areas. These intervals vary with many factors such as type of crop or tree, wind, rain, and specific phosphate compound used.

These phosphate poisons enter the body rapidly and easily by ingestion, inhalation and absorption through intact skin. A distinct danger lies in the fact that signs and symptoms of illness do not appear until dangerous quantities have been absorbed. Subclinical poisoning may exist from chronic exposure, then acute illness develop suddenly and progress swiftly upon relatively small additional exposure.

These manifestations result from excessive cholinergic stimulation and are typically:

• Muscarinic-like symptoms. Symptoms referable to the gastrointestinal tract are vomiting, cramping, eructation, diarrhea, tenesmus and rectal incontinence. Respiratory tract involvement is manifest by tightness, wheezing, increased bronchial secretions, cough, dyspnea and subsequent pulmonary edema with cyanosis. Symptoms referable to

the urinary tract may include frequency and incontinence. Other symptoms of poisoning of the muscarinic type are sweating, lacrimation, salivation, myosis, blurring of the vision and bradycardia.

- Nicotinic-like poisoning manifestations of twitching, cramping, weakness, fasciculation of skeletal muscles and respiratory depression. Also, if the sympathetic ganglia are affected, pallor and blood pressure elevation may be manifest.
- Central nervous system manifestations include giddiness, nightmares, confusion, ataxia, convulsions, coma and depression of respiratory and circulatory centers.[9]

LABORATORY FINDINGS

Diagnostic laboratory evidence of phosphate poisoning is obtained by measuring erythrocyte cholinesterase levels. Many procedures have been devised for this determination.[3] Additionally, a serum cholinesterase level measurement has been used in this regard but is nonspecific because serum cholinesterase is inactive, is affected by numerous other factors and has consequently been designated as a *pseudo*cholinesterase. Materials and instructions for a readily available, easily performed test that can be used in screening for possible chronic poisoning as well as for confirmation of an acute clinical situation are obtainable from the Biological Test Products Corporation, 1112 Thompson Avenue, Rozelle, New Jersey. This method tests whole blood acetylcholinesterase levels and provides all information needed. The authors often have observed leukocytosis with neutrophilia in patients with phosphate poisoning. Transitory albuminuria has been noted, but not hematuria. There are no other diagnostic laboratory findings.

ANTIDOTES

An ideal antidote would be one which competed with the phosphates for the binding area of the acetylcholinesterase and which also dephosphorylated the inactivated enzyme. If such an antidote were only competitive for the binding area, one would expect eventual restoration of the acetylcholinesterase. There is reason to believe that this would take some time, as judged by the length of time it takes for the erythrocyte acetylcholinesterase to return to normal after withdrawal from exposure to poison. Therefore, such an antidote presumably would not be immediately effective. However, an antidote which allows dephosphorylation and thereby restores the capability of hydrolysis could potentially result in immediate reversal of the poisoning mechanism. Substantial evidence has been accumulated that one compound, called pyridine-2-aldoxime methiodide, and known as PAM, achieves both of the above-mentioned antidotal properties.[2] In clinical usage, full restoration of acetylcholinesterase to normal levels is not readily and continually achieved although the condition of the patient shows rapid clinical improvement.

It should be stressed that inhibition of erythrocyte acetylcholinesterase activity may persist for several months after withdrawal from further exposure. Decreased serum acetylcholinesterase activity ascribable to poison may last for weeks, and in this regard it should be pointed out that there is varying toxicity between different forms of the phosphate poisons. The type known as Parathion is considerably more toxic than Malathion. Parathion is converted in the body to a substance called paraoxon, which is the actual cholinesterase inhibitor. The rate of excretion of PAM is faster than the conversion of Parathion to paraoxon, so symptoms may recur after initial treatment with PAM and subsequent therapy may be necessary in any given case.[8]

Atropine was the first practical remedy because of some counteracting effects of acetylcholine excess. Its inadequacy as a complete antidote stems from the fact that although it reverses the muscarinic effects well, it does not reverse the nicotinic effects or for the most part the central nervous system effects of acetylcholine. With atropine, other measures such as control of convulsions with barbiturates and the use of supportive means such as artificial respiration were often necessary. Substantial numbers of reports of the use of PAM in phosphate poisoning have been published, particularly in the foreign literature. The paucity of similar reports in the American literature is interesting. The *Journal of the American Medical Association* in 1958 printed an article from Japan reporting on administration of PAM intravenously. The signs of poisoning were frequently reversed in a very short period.[6] Rapid clearing of the nicotinic and central nervous system effects have been reported by many investigators. There is, however, some uncertainty as to whether PAM counteracts in full or part the muscarinic manifestations of excess acetylcholine. For this reason, it is probably advisable to use both drugs in a severely poisoned person. The clear superiority of PAM over atropine in overall effect, however, is shown in many case reports and is documented in one of the cases reported herein. The duration of PAM effect is apparently relatively transient, and repeated doses may be necessary. This is a limiting factor in the potential of the drug as a prophylactic agent.[9] Side effects have not been reported in clinical use. The $L.D._{50}$* of PAM in mice is variably reported as 110 to 190 mg. per kg. of body weight. PAM can be given at the rate of 500 mg. per minute intra-

*Lethal dose for 50 per cent of subjects.

venously. The crystalline material is 5 per cent soluble so that the methiodide form in solution has 1 gram per 20 cc. of water.

PAM is considered experimental and if it is used in a new form called Protopam chloride, one must have the patient's permission to give it as an experimental drug and the physician must submit a Statement of Investigation. The drug is at present available under the above conditions through the Campbell Pharmaceuticals, Inc. The substance has been demonstrated to reactivate 80 per cent of phosphate-inhibited enzyme within one minute.[4] Animal studies have shown the duration of protection dropping substantially after the first hour. Some of the cholinesterase results noted in the cases here reported tend to show the same phenomenon in humans.

REPORTS OF CASES

CASE 1. An 18-year-old boy who spent his summer vacation loading Parathion insecticides into airplanes for use in crop dusting was admitted to hospital late at night. He stated he had frequently been exposed to the chemical, both by inhalation and by spilling it upon himself. On the afternoon of the day of admission he noted rather rapid increasing weakness, abdominal cramping, pain, nausea and muscle twitching. He denied increased cough, perspiring or salivation. He lay limp and quiet, appeared weak and mentally torpid. He complained of abdominal distress and frequently gagged but did not vomit. His skin was moist. Respirations were 20 per minute and the pulse rate 84 (an hour later 72). Blood pressure at first was 160/80 mm. of mercury and later 130/75. The pupils were small but not pinpoint. Atropine 0.45 mg. had been administered 15 minutes before his admission. Increased salivation was noted. The abdominal muscles seemed taut in comparison with frail, weak muscles elsewhere. No increased rales or rhonchi were heard. Deep tendon reflexes were decreased to absent but symmetrical. Testing of muscles was estimated to reveal 70 per cent decreased strength symmetrically. The screening acetylcholinesterase depression test confirmed the admitting diagnosis. We had not previously used PAM (this was in August 1960) although we had obtained a supply and prepared it by placing it in solution and sterilizing it.

Blood was drawn for laboratory studies, including the screening test for cholinesterase level as well as for more accurate determination of serum and erythrocyte levels to be done subsequently. At this point, 0.75 gm. of PAM was given slowly intravenously. In a matter of minutes, the abdominal cramping cleared, the muscle cramping and weakness eased and the patient appeared a good deal stronger, which he was, as tests showed. The nausea had passed. He remained quite comfortable under close observation throughout the night, and the following morning the pupils were normal and there was no longer any muscle discomfort, weakness or other sign of poisoning. That subacute or chronic poisoning effects persisted, however, was indicated by the levels of anticholinesterase in the blood. Laboratory data on acetylcholinesterase contents are shown in Table 1. At the time of admission, leukocytes numbered 21,000 per cu. mm., with a cell differential of 89 per cent neutrophils, 8 per cent stabs, 2 per cent lymphocytes and 1 per cent metamyelocytes. The carbon dioxide combining power was 27 milliequivalents. The following day, leukocytes numbered 11,700 with normal cell differential. Except for a heavy trace of albumin, results of urinalysis were within normal limits.

CASE 2. A 20-year-old man working for the same crop dusting concern as the patient in Case 1 had been handling concentrated Parathion material in loading tanks for some two months. The history was obtained from the patient, the referring physician, and persons at the place of employment. Not all the information was available before treatment. Approximately seven to ten days before admission, the patient had spilled Parathion solution on his clothing, wetting his chest and abdomen and legs. He had immediately showered and apparently had had no immediate symptoms. At 10 a.m. the day of admission, he spilled a small quantity of the poison in liquid form of unknown concentration on his right foot. He washed his foot and canvas shoe with plain water but then continued to wear the shoe after this inadequate cleansing. At about 2:30 p.m., nausea developed and was soon followed by sweating, weakness and excessive salivation. At this time the patient took two 0.4 mg. tablets of atropine. He subsequently vomited and was then admitted to hospital. At this time his principal complaint was of severe trembling of the muscles of the arms, neck and upper trunk.

Upon physical examination the patient appeared to be acutely ill and the skin was cold and sweaty. Respirations were normal. Considerable salivation was present and the speech was thick and slurred. The patient was irritable, asking to be let alone rather than examined. The most striking physical finding was a tremendous play of fasciculations of the muscles, especially of the arms, shoulder girdle and neck. The neck was supple. The pupils were at normal positions, and were about 3 or 4 mm. in size. At first the pulse rate was 64 and after 20 minutes at the emergency room had become 48. Blood pressure was 160/90 mm. of mercury. Respirations subsequently became mildly depressed. Trembling fasciculations made deep tendon reflex testing difficult, but good ankle jerks were obtainable although the knee reflexes were not elicited. At 4:20 p.m., 1.2 mg.

of atropine was given subcutaneously. There was no effect on the symptoms. At 5:00 p.m. the same dose of atropine was given intravenously over a period of two minutes. There was immediate flushing of the skin and, within minutes, complete cessation of the profuse perspiration. From then on the patient's skin was warm and dry. The heart rate increased from approximately 50 to 140, and tachypnea with shallow, rapid respirations ensued. Rhonchi and excessive salivation cleared completely. Blood pressure was unchanged as were the pupils and the tremor. The patient remained weak. At approximately 5:30 p.m., although the eyes were open and the patient seemed to see and comprehend, he did not speak or respond. At 5:45 p.m., the first of four grand mal seizures over a period of 50 minutes occurred, the first lasting 15 minutes, the second 9 and the last two 6 minutes each. It appeared that atropine had accomplished all that it could, in that there was complete clearing of the muscarinic effects of acetylcholine, since excess oral and tracheobronchial secretions had cleared, and the skin was perfectly dry. Because of this and the rapid heart rate, undesirable effects of the usually advised, higher doses of atropine were feared. The nicotinic and central nervous system effects of severe phosphate poisoning had not been affected. Progressive central nervous system deterioration was present. Immediately after the first convulsion, sodium luminal was given intravenously, and sodium amytal intravenously with the second convulsion. PAM was given during the fourth convulsion, 0.5 gm. slowly by vein over a period of three minutes. As the patient relaxed from this convulsion, the fasciculations of muscles, which had continued throughout and which were quite pronounced, ceased completely and did not recur. Respirations had become depressed and the heart rate, which had been 140, continued at that rate. The pupils had not dilated and were essentially normal in position. At 7:15 p.m., a second dose of PAM was given intravenously and within five minutes or so respiration was normal. At 8:20 p.m., the patient aroused to consciousness, having been unconscious for about two hours and 50 minutes. By 10:00 p.m., he was alert, cooperative and gave a full history, although he was still weak. The following morning, he was well without complaint except for dryness of the mouth because of continued atropine. No weakness was demonstrated. The cholinesterase screening test indicated marked inhibition, and the following day the test indicated complete inhibition. Thirteen days later, the screening test again showed marked inhibition. There had been no further exposure. Results of cholinesterase determination are shown in Table 2.

CASE 3. The patient, the same as the one in Case 1, was poisoned again a little over a year later.

TABLE 1.—*Results of Cholinesterase Determination, Case 1*

Time	Whole Blood Cholinesterase Testing	Plasma	Erythrocytes
Initial (before PAM)	*Complete	*0.0 delta ph	0.18 delta ph
10 minutes (after PAM)	Marked	0.27 delta ph	0.44 delta ph
45 minutes (after PAM)	Moderate	0.10 delta ph	0.44 delta ph
Next a.m. (10 hours)	.....	0.0 delta ph	0.31 delta ph

Normals:*
Whole Blood = No inhibition.
Plasma 0.41—1.65 delta ph
Erythrocyte 0.55—1.25 delta ph

*The results of the screening test (whole blood) that we use for cholinesterase activity are reported in four categories: No inhibition, moderate inhibition, marked inhibition, complete inhibition.

TABLE 2.—*Results of Cholinesterase Determination, Case 2*

Time	Erythrocytes		Plasma	
	Delta ph	Per Cent of Published Normals	Delta ph	Per Cent of Published Normals
60-90 minutes after PAM	.40	54	.17	23
14 hours	.30	40	.08	11
38 hours	.31	41	.19	27
18 days	.37	49	.48	68

Again he had spent his summer working in loading airplanes for crop dusting. He believed that he might have spilled Parathion on himself two weeks before admission to hospital. Then, two nights befor entering the hospital, a high pressure hose that was being used to load a plane parted at a coupling and fluid spilled, drenching the patient's clothing below the abdomen. He immediately jumped into an irrigation ditch and washed himself thoroughly. He did not change his clothes, however, and continued to work. Except for being a little tired, he had had no symptoms the day before admission. Then on the afternoon of the day he entered the hospital, while working, he noticed gradually increasing muscle weakness and excessive perspiration. This was followed by nausea and vomiting. He was able to get home but because of extreme muscle weakness, he had to be helped into the shower. Muscle jerking was noted but no fibrillary twitching. Vision was not abnormal. The patient did not lose consciousness. At the time of admission, his skin was wet, the pupils normal, and he was lying on the bed retching. Involuntary muscle twitching, but without constant or repetitive fibrillation, was noted. PAM was administered slowly by vein, 1 gm. in 20 cc. of water. Nausea continued for about 30 minutes, but within an hour all nausea and muscle weakness were gone. The following morning, the patient felt well and no abnormalities of physical signs were detected. In this case the initial screening test for acetylcholi-

nesterase activity, carried out 30 minutes after the administration of PAM, showed marked inhibition. The cholinesterase screening test the following morning showed complete inhibition, and a test obtained nine days later, again after no further exposure, showed marked inhibition of the total blood level.

The authors have observed two cases in addition to those here reported in which the patient (in one instance a 22-month-old child) spilled or swallowed Malathion. The use of PAM was not necessary. Also, two other instances of agricultural poisoning have recently occurred in our community, and in both instances PAM was used intravenously. In these cases the patients were treated by other physicians with supervision by the authors. Prompt response was noted. Laboratory data are not available.

DISCUSSION

It has been our feeling after a substantial review of this subject, that further education in regards to the handling of these phosphate poisonings is indicated. Worker education, protection and instructions in many fields is necessary. In our area in the Sacramento Valley, there are numerous agricultural processes whereby exposure can be achieved. Studies to determine the duration and type of exposure necessary for clinical poisoning or dangerous levels are probably indicated. An excellent article in this regard demonstrates the problems presented in greenhouse exposure to these poisons.[1]

Periodic acetylcholinesterase levels on persons with industrial or agricultural exposure is probably indicated. Our area has many people engaged in airplane crop dusting with these phosphate poisons. Ground personnel, including those who signal the airplanes as well as those who load and handle the poisons, are frequently exposed. One group has been under our observation with periodic cholinesterase determinations. Two members of this group have shown moderate inhibition by the screening test and possibly serious inhibition and poisoning was prevented by effecting withdrawal from exposure. Corroborative red blood cell acetylcholinesterase levels were obtained in each case and were confirmatory of inhibition.

Labeling of insecticides with substantial warning is important. We have seen poisoning in youngsters where little warning was given to the parents of the possible dangers. Likewise, identification of the poison, by the physician, has not always been easy. Repeated cases of poisoning that we have seen have come from one crop dusting concern where education and caution have not been observed, notwithstanding our repeated admonitions along this line.

Governmental supervision and law enforcement at federal, state, county and local levels has not, in our experience, been adequate. The departments assigned to these tasks are substantially understaffed in relation to the widespread use of these poisons. Three branches of our state government are concerned with different phases of control of these poisons and a unified approach has not been achieved.

Better physician education is equally important. A prime example is that queries directed to some poison control centers result in information concerning only atropine as an antidote for phosphate poisoning. Even pamphlets distributed at the present time by the California State Department of Health make no mention of PAM. An article in this month's (October 1961) issue of the journal, *G.P.*, likewise does not mention PAM.[7]

How much morbidity and how many fatalities will ensue before proper education of all concerned is achieved? How long will this life saving antidote be available only as an "experimental investigator" drug?

It is to be noted in Tables 1 and 2 that substantial correction of cholinesterase inhibition does not occur though rapid clinical improvement is achieved by the antidote.

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